

## **CLAIMS**

What is claimed is:

1. A method for performing interactions, using a microscope, on microscopic subjects that change in space and time, comprises the following steps:
  - acquiring at least one image of a sample that encompasses at least one microscopic subject;
  - defining by the user virtual reference subjects on a discrete grid of the acquired image or images, in order to define regions;
  - automatic acquiring of a sequence of image data or volume data;
  - successive identifying an optical flux based on the sequence of acquired images;
  - applying the identified optical flux to the defined reference subjects; and
  - performing interactions on the reference subject modified by the optical flux.
2. The method as defined in Claim 1, wherein the virtual reference subjects are defined as regions, discrete point sets, or local coordinate systems on the reference grid of the image sequence, and define interaction locations.
3. The method as defined in Claim 1, wherein the interactions encompass the recovery of measured data of the subject or the controlled manipulation of the subject at the positions defined by the virtual reference subjects.
4. The method as defined in Claim 3, wherein the recovery of the measured data encompasses
  - image data;

- geometrical data of the virtual reference subject, such as center point, area, periphery, or volume, or
- magnitudes derived from geometrical data of the virtual reference subject, such as velocity, acceleration, volume and area growth rates, and collision statistics of virtual reference subjects; and
- determination of the intensity within the region defined by the virtual reference subject, by acquisition of the intensity distribution function and any desired parameters derived therefrom, such as mean, variance, skewness, or higher elements, as well as other parameters common in statistics such as quantile, median, or range width.

5.

6. The method as defined in Claim 3, wherein the controlled manipulation of the subject is accomplished with respect to the specific regions or positions in the subject by means of radiation, for purposes of bleaching, photo activation, cage-compound release, and cutting and excision.

6.

A system for interactions on microscopic subjects that change in space and time comprising:

- a confocal scanning microscope that guides an illuminating light beam over a subject;
- several detectors that identify, from the light proceeding from the subject, intensities from different spectral regions;
- a processing unit;
- a PC;
- an input unit;

a display on which an individual image is presented to the user; the user interactively defines virtual reference subjects on the image shown on the display, using the input unit for position definition;

a means for determining the optical flux based on the intensities from different spectral regions identified by the detectors is housed in the processing unit; and

a means for applying the optical flux to the virtual reference subjects is present in the processing unit and the processing unit controls interactions on the basis of the changed reference subjects.

7. <sup>4</sup>  
8. The system as defined in Claim 7, wherein the virtual reference subjects are defined as regions, discrete point sets, or local coordinate systems on the reference grid of the image sequence, and define interaction locations.

8. <sup>4</sup>  
9. The system as defined in Claim 7, wherein the interactions accomplishes both the recovery of measured data of the subject and the controlled manipulation of the subject at the position defined by the virtual reference subjects.

9. <sup>8</sup>  
10. The system as defined in Claim 9, wherein the recovery of the measured data encompasses

- geometrical data such as center point, area, periphery, or volume, or
- magnitudes derived from geometrical data, such as velocity, acceleration, volume and area growth rates, and collision statistics of virtual reference subjects; and
- determination of the intensity within the region defined by the virtual reference subject, by acquisition of the intensity distribution function and any desired parameters derived therefrom, such as mean, variance, skewness, or higher

elements, as well as other parameters common in statistics such as quantile, median, or range width.

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H. The system as defined in Claim 10, wherein the controlled manipulation of the subject is accomplished with respect to the specific regions or positions in the subject, the manipulation by means of radiation, for purposes of bleaching, photoactivation, cage-compound release, and cutting and excision.